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Zoonotic infections among veterinarians in Turkey: Crimean-Congo hemorrhagic fever and beyond

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Summary
Objectives: We aimed to determine the seroprevalence of Crimean-Congo hemorrhagic fever (CCHF) virus, *Brucella spp*, and *Coxiella burnetii* among veterinarians in a highly endemic and a non-endemic region for these infections in Turkey.
Methods: The antibody levels against these three infections were surveyed. Eighty-three veterinarians were included from two distinct geographic regions.
Results: CCHF IgG positivity (3% vs. 0%) and Brucella agglutination titer of $\geq$1/160 (33% vs. 5%) were more common in the endemic region, whereas the rates of *Coxiella burnetii* antibodies were similar (7% and 8%). A history of tick bite was significantly more common in the endemic region (35% vs. 12%, $p = 0.011$). A multivariate analysis was performed among the veterinarians living in the endemic area, and percutaneous injuries were found to be associated with Brucella infection (OR 1.8, CI 1.09—3, $p = 0.022$).
Conclusions: Veterinarians should protect themselves against tick bites, and should use masks to prevent transmission by inhalation of zoonotic infections in endemic countries.

Introduction

Zoonotic infections are potential occupational hazards among veterinarians and agricultural workers. The recent emergence of Crimean-Congo hemorrhagic fever (CCHF) infection in the Tokat region of Turkey,[1,2] inspired us to study

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the seroprevalence of Crimean-Congo hemorrhagic fever virus (CCHFV) infection among one of the leading risk groups, veterinarians. CCHF is a disease reported in Africa, the Middle East, some parts of Southern Europe, Russia, and China. The virus from the *Nairovirus* genus, Bunyaviridae family causes severe diseases in humans, with a mortality rate of 5—30%.[3—6] Humans become infected mostly through tick bites or direct contact with body fluids or tissues from viremic patients or viremic livestock.[5—7] Since some of the symptoms of CCHFV might mimic Brucella and Coxiella infections, the antibodies against these two zoonotic infections were also surveyed.

Brucellosis is an endemic zoonotic infection in Turkey, and has been reported to be common among cattle and sheep in Tokat, whereas not in Aydin.[8] In transmission of *Brucella spp*, contact with infected animals or animal products is important. Workers in the dairy industry, shepherds, farm workers, family members who have contact with animals around the home, abattoir workers, kitchen workers, and veterinarians are all at risk of infection. Cuts and abrasions on the hands and forearms are sites of entry of infected material. Aerosols of infected fluid are also sources of infection, and entry of organisms may take place across mucosal surfaces.[9]

Q fever is usually an occupational disease affecting those with direct contact with infected animals, such as farmers, veterinarians, and abattoir workers. Humans may become infected by inhalation of small-particle aerosols containing *Coxiella burnetii*.[10] Performing deliveries or curettages are the leading risk factors for infection. *C. burnetii* infection has previously been reported from Tokat, but not from Aydin.[11]

We studied the seroprevalence of antibodies against these common zoonotic infections in Turkey, and analyzed the risk factors.

Methods

Subjects

The study was conducted in 2003 in two distinct regions of Turkey, after a CCHF outbreak in the eastern part of the country. The study group included veterinarians from an endemic province (Tokat) and a non-endemic province (Aydin) of Turkey as shown on the map (Figure 1). Tokat had the highest number of human CCHF cases in the 2002—2003 epidemic of CCHF, whereas Aydin has the lowest rate of zoonotic infections in Turkey.

A structured survey was administered to obtain information about the risk factors of zoonotic infections among veterinarians during the same time period in both regions. The data on demographics, the length of professional experience, the number of deliveries and curettages performed within the previous 6 months, the number of percutaneous injuries within the previous 6 months, and the rate of adherence to personal protective equipment use (gloves, gown, goggles, and mask) were obtained. Additional information was collected regarding any clinical signs or symptoms among veterinarians during the previous six months, including fever, myalgia, headache, history of tick bite and history of consuming raw milk or fresh cheese. Informed individual consent was obtained from the veterinarians participating in the study.

Serologic studies

The sera from the veterinarians were collected one month after the last case in the hospital (October 2003), in the same time period for both regions. Specific CCHF IgM and IgG antibodies were studied by enzyme-linked immunosorbent assay (ELISA). The presence of IgG against *C. burnetii* phase II

Figure 1 Endemic (Tokat) and non-endemic (Aydin) provinces on the map of Turkey.

Table 1 Personal and professional characteristics of the 83 veterinarians

Age, median (min—max) years	35 (23—50)
Male gender	76 (92%)
Length of professional experience, median (min—max) years	12 (1—24)
Number of deliveries performed within the previous 6 months, median (min—max)	11 (0—100)
Number of curettages performed within the previous 6 months, median (min—max)	10 (0—200)
Veterinarians with a history of tick bite	19 (23%)
Percutaneous injuries within the previous 6 months, median (min—max)	2 (0—5)
The rate of adherence to protective measures during work	
Boots	70 (84%)
Gloves	69 (83%)
Gown	68 (82%)
Mask	9 (11%)
Goggles	10 (12%)

was measured using commercially available indirect immunofluorescent antibody (IFA) (Vircell SL®, Granada, Spain). According to the manufacturer's recommendation a titer of at least 1/64 was considered positive. Anti-*Brucella spp* antibodies were determined by Brucella microagglutination test (MAT), as described previously.[12] Brucella agglutination with a titer of ≥160 was defined as positive.

Data analysis

Data were analyzed using Stata Statistical Software, version 8.0 (Stata Corporation, Texas, USA). Mean comparisons for continuous variables were done using independent groups *t*-tests. Proportion comparisons for categorical variables were done using Chi-square tests, although Fisher's exact test was used when data were sparse. Univariate and multivariate analyses were performed to detect the risk factors associated with Brucella and Coxiella infections separately. The association of age, gender, the length of professional experience, the number of deliveries and curettages performed within the previous six months, the number of percutaneous injuries within the previous six months, and the rate of adherence to personal protective equipment use (gloves, gown, goggles, and mask) were tested. The analyses were repeated for the endemic and non-endemic region, separately. The statistical significance was set as p value of <0.05.

Results

Eighty-three veterinarians, 40 from an endemic region (Tokat), and 43 from a non-endemic region (Aydin) were enrolled in the study (Figure 1). Eighty percent of the veterinarians in the endemic region, and 20% of the veterinarians from the non-endemic region were included. All the veterinarians had cared for cattle and sheep. The characteristics of veterinarians from both regions were similar in terms of their age ($p = 0.366$), gender ($p = 0.768$), and the length of professional experience ($p = 0.765$). However, the veterinarians from the non-endemic region performed more professional activities such as deliveries ($p < 0.001$) and curettages ($p = 0.001$) than their colleagues from the endemic region.

A history of tick bite was significantly more common among the veterinarians from the endemic region (35% vs. 12%, $p = 0.011$). Seventy percent of the veterinarians had a history of percutaneous injury, and the median number of injuries was two within the previous six months (from April 2003 to September 2003, Table 1). None of the veterinarians had a history of consuming raw milk or fresh cheese. The rate of adherence to the use of boots, gowns, and gloves was higher than that for the use of masks and goggles (Table 1).

Thirteen veterinarians in the endemic region and two in the non-endemic region had a Brucella agglutination titer of ≥160 (Table 2). Among these, four veterinarians in the endemic region had symptoms of fever, malaise, and myalgia within the previous six months, which were compatible with Brucella infection. In a multivariate analysis, the veterinarians living in the endemic region were found to have a higher rate of Brucella infection than those living in the non-endemic area. The sub-group analysis of risk factors for Brucella infection in the endemic region revealed that the veterinarians who had a higher rate of percutaneous injuries had higher Brucella agglutination titers (OR 1.8, CI 1.09—3, $p = 0.022$).

In one veterinarian from the endemic region, CCHF IgG antibodies were found to be positive. His Brucella agglutination titer was 1/160. He had no history of tick bite within the previous six months, but had had five subsequent percutaneous injuries. Within this period he had had symptoms of malaise, fever, and fatigue, which were compatible with both brucellosis and the milder form of CCHF infection. He did not have any biochemical tests at that time.

The prevalence of *Coxiella burnetii* serology was almost equal in both regions, and the seroprevalence was six out of 83 (7%); none of them had any complaints (Table 2).

Table 2 Serologic results versus history of symptoms (malaise, myalgia, and fever) within the previous six months

	Endemic region $N = 40$ (%)		Non-endemic region $N = 43$ (%)	
	Serology	Symptoms	Serology	Symptoms
CCHF IgG (ELISA)	1 (3)	1 (3)	0	—
Coxiella burnetii IgG (IFA)	3 (8)	—	3 (7)	—
Brucella agglutination ≥160	13 (33)	4 (10)	2 (5)	—

CCHF, Crimean-Congo hemorrhagic fever; IFA, immunofluorescent antibody.

Discussion

The zoonotic infections could have common mechanisms of transmission, such as percutaneous injuries, handling of animals, or inhalation. However, the infectivity rate of each microorganism differs, and some transmission routes are more significant for some infections. Therefore, the risk factors for each zoonotic infection should be evaluated separately.

CCHF infection is not symptomatic in animals. However, besides contact with ticks, handling and slaughtering viremic domestic animals have been reported to be important risk factors for CCHF.[6] In a study from South Africa among 484 farm workers, CCHF antibody prevalence was found to increase with age, and was correlated with the handling of sheep.[13] In another study, the prevalence of antibodies against CCHF was found to be greatest in large mammals, which are known to be the preferred hosts of the adult tick (Hyalomma) vectors of the virus.[14] In our study, a history of tick bite was significantly more common in the endemic region than the non-endemic region (35% versus 12%, $p = 0.011$). Within the CCHF outbreak season, 14 veterinarians in the endemic region (35%) had a history of tick bite, and CCHF IgG was detected only in one veterinarian, who had no history of tick bite. He might not have noticed the tick bite, or he might have acquired the infection through his percutaneous injuries. Among CCHF patients, 53% reported tick bites.[1]

Besides CCHFV infection, brucellosis was also highly endemic in Tokat region. Brucella infection among veterinarians has been reported from Eritrea (4.5%)[15] and Lebanon.[16] A higher rate of seroprevalence has been detected in the USA (17.8%)[17] and northern Jordan.[18] In our study Brucella agglutination against Brucella was found to be very high among veterinarians from the endemic region (33%), and 10% of the veterinarians had symptoms and signs compatible with brucellosis (Table 2). A study from the USA reported that 13.9% of veterinarians had a history of a prior clinical illness diagnosed as brucellosis.[17] Our study shows that the rates of adherence to protective measures, particularly the use masks and goggles were very low (Table 1). None of the Brucella seropositive veterinarians used masks while they were working. Transmission of the infection by inhalation among veterinarians could be underestimated as happened in hospital settings in Turkey.[19] The higher rate of percutaneous injuries was another significant risk factor (OR 1.8, CI 1.09—3, $p = 0.022$). Our results should alert the authorities to implement effective preventive measures.

Q fever infection has previously been described in occupational risk groups, including veterinarians and agricultural workers.[20—28] In sero-epidemiologic studies among veterinarians, phase II *C. burnetii* IgG antibodies were found in 13.5% in Japan,[23] 17% in Australia,[24] 84% in the Netherlands,[25] and 13% in Sweden,[26] which were higher than in the normal population. The rate of *C. burnetii* seropositivity was detected at around 7—8% of the general population and the veterinarians in Turkey.[27,28] Our finding of 7.5% *C. burnetii* seropositivity in both regions suggests that *C. burnetii* infection is not common in Tokat region, in contrast to other infections that have been studied. Furthermore, *C. burnetii* seropositivity among veterinarians was not found to be higher than in the general population, as has been indicated in previous studies.[27,28]

Our study group represents 80% of the veterinarians in the endemic region and 20% of the veterinarians in the non-endemic region. However, this proportion of the veterinarians in the non-endemic region was actively working with cattle and sheep, and the remaining veterinarians were working in chicken farms.

CCHF and brucellosis are the leading occupational infection risks for veterinarians in the endemic regions. Veterinarians should be warned to protect themselves against tick bites. The use of masks and goggles should be employed to prevent transmission of CCHF and brucellosis.

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Conflict of interest: No conflict of interest to declare.

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